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Original Article

Diagnostic and prognostic value of CXCL12 (SDF-1α) level in *Mycobacterium tuberculosis* infection and disease

Ilkay Koca Kalkan[1], Ayşe Gozu[2], Ebru Tansel[2], Serife Nilgun Kalac[2], Belgin Samurkasoglu[2], Hulya Simsek[3]

[1] Department of Allergy and Clinical Immunology, Ataturk Chest Disease and Thoracic Surgery Training and Research Hospital, Ankara, Turkey
[2] Department of Pulmonology, Ataturk Chest Disease and Thoracic Surgery Training and Research Hospital, Ankara, Turkey
[3] Department of Medical Microbiology, Yozgat Bozok University Faculty of Medicine, Yozgat, Turkey

Abstract

Introduction: New diagnostic tools are being investigated for rapid and accurate TB detection. We aimed to find out the diagnostic yield and accuracy of chemokine CXCL12 (SDF-1α) levels in diagnosing active TB (aTB) and making a differential diagnosis from other several infectious/non-infectious pulmonary conditions.

Methodology: We collected demographic, clinic features and studied plasma CXCL12 levels using ELISA kit of the participants, classified into five categories: aTB (n = 30); cured TB (cTB, n = 15); close contacts of aTB (CC, n = 15); chronic obstructive pulmonary disease (COPD) with active nonspecific pulmonary infection (infCOPD, n = 15); and healthy controls (HC, n = 15).

Results: CXCL12 levels were highest in aTB, but no significant difference was seen between other groups. When a cut-off level for CXCL12 was determined as 2835 pg/mL, the increased CXCL12 rate was significantly more in aTB than CC and HC ($p = 0.02$, $p = 0.05$). Also, participants with an active infection (aTB and infCOPD) had significantly higher increased CXCL12 rates ($p = 0.01$). The sensitivity and specificity of CXCL12 for diagnosing aTB were found to be 0.56 and 0.63, respectively. We found that bacterial load, the radiological severity and the extent of chest x-ray involvement were independent factors for increased CXCL12 levels.

Conclusions: Our study demonstrates that CXCL12 may be a representative of active pulmonary infection regardless of the cause but correlated with the severity of the disease; enabling this test to be used as a prognostic factor rather than a diagnostic test for aTB.

Key words: CCXCL12 (SDF-1α); COPD; diagnostic test; infection; prognostic test; tuberculosis.

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Introduction

As a global infection, tuberculosis (TB) is a communicable disease with an estimated 10.0 million new cases in 2018, a number that has been relatively stable in recent years. It is one of the top 10 causes of death worldwide. Since 2011, it is the leading cause of death from a single infectious agent (surpassing HIV). Only in 2018 1.5 million people died from TB. Its incidence is 132/100.000 due to data of the 2019 World Health Organization (WHO) reports [1].

The END TB strategy aims to reduce TB incidence and mortality in 2025 (compared to 2015 figures) by 50%, and 75%, respectively [2]. In achieving these ambitious targets, related factors that underlie and contribute should be improved; like reducing the delays in health care seeking, diagnosis and treatment. Early diagnosis improves survival by identifying cases with TB in order to prevent further transmission and to give effective treatment [3].

A large proportion of TB mortality occurs in those who have not been diagnosed. Smear microscopy, which is the most frequently used test for TB diagnosis in the last 100 years, has poor sensitivity. The current gold standard tests to diagnose active TB are still the microbiological tests with the disadvantages of being insensitive or time-consuming [3]. Globally, more than 2000 million people are estimated to be infected with *Mycobacterium tuberculosis* (M.tbc), but only 5-15% ever develop active TB. It is important to identify and differentiate latent from active TB [2]. T-cell based IFN-γ release assays (IGRAs) cannot distinguish active TB from latent TB [4]. The only rapid diagnostic test for the detection of active TB and rifampicin resistance, recommended by WHO, is the Xpert MTB/RIF® assay,

which is expensive and raises concerns about its accessibility in low- and middle-income settings [3]. Several autopsy-based studies have shown that diagnosis is often missed in patients with smear-negative tuberculosis, disseminated TB and extrapulmonary TB. Also, in high-TB incidence and high HIV prevalence settings smear-negative disease is one of the major risk factors for mortality [5-10]. The availability of point-of-care diagnostics is an important intervention that can reduce TB mortality [1]. New diagnostic tools are thus required for rapid and accurate detection of TB urgently.

After *M. tuberculosis* infection, both innate and adaptive immunity play an important role in recruitment/activation of T lymphocytes (predominantly T helper 1 (Th1)), monocyte and macrophages resulting in granuloma formation at the site of infection [11]. This caseating granuloma, the histologic hallmark of TB, keeps the infection within and hedge rounds passing in and out of the tissue.

Chemokines are a large superfamily of tiny molecular peptide molecules (8-14 kDa) that promote migration and adhesion of various types of leukocytes to areas of injury and functionally play different roles in both inflammatory and homeostatic processes [12,13]. The CXC chemokine CXCL12, also known as stromal cell-derived factor-1α (SDF-1α), is a widely expressed chemokine, including most leukocytes. It has a broad range of actions that one of which is controlling the trafficking of lymphocytes [12]. Since Th1 lymphocyte is known to play a central role in anti-tuberculosis immune response and CXCL12 is one of the most efficacious T lymphocyte chemoattractants and activators, CXCL12 may be a potential biological marker for TB diagnosis. There is limited but promising data concerning the diagnostic value of this chemokine for active tuberculosis infection [l,4,14,15]. However, there may be other infections that can also trigger mobilization and accumulation of T lymphocytes by inducing chemokines like CXCL12.

In this study we aimed to find out the diagnostic yield and accuracy of serum CXCL12 / SDF-1α levels in diagnosing active TB, and making a differential diagnosis from other several infectious/non-infectious pulmonary conditions.

Methodology

Study Population

This study was carried out at one of the largest tertiary referral pulmonology hospitals in the country. The study protocol had been approved by the institutional ethical committee. A written informed consent form was obtained from all participants. All procedures performed in the current research were in accordance with the Declaration of Helsinki. All subjects were 16 years and over.

All the participants enrolled were classified into the following five categories: (1) 30 patients with active TB (aTB); (2) 15 patients with cured TB (cTB); (3) 15 participants who are close contacts of aTB patients (CC); (4) 15 patients with chronic obstructive pulmonary disease (COPD) with active nonspesific pulmonary infection (infCOPD); and (5) 15 healthy controls (HC).

Definition of Study Groups

Active TB patients were the ones who had confirmed disease microbiologically by smear and/or culture positivity for *M.tbc*. No aTB patients had started treatment at the time of enrolment. Cured TB patients had proved sputum smear conversion from positive to negative at least two times, one during treatment and the other at the completion of therapy. Close contacts of aTB patients without any clinical, radiographic or microbiologic evidence of TB were classified as CC. Patients labeled as infCOPD are the ones who had an acute exacerbation of COPD caused by respiratory tract infections, with new onset respiratory symptoms (e.g. cough and sputum) and inflammatory findings (e.g. leucocytosis, fever and increased serum level of erythrocyte sedimentation rate) in the absence of acid-fast bacteria and abnormal chest X-ray shadows. Volunteers without any pulmonary symptoms or active disease were recruited as HC.

Exclusion Criteria

We excluded patients with chronic diseases as rheumatoid arthritis, diabetes mellitus, acute hepatitis or liver cirrhosis, malignant tumours and immunosuppressed conditions (including HIV infection) that can cause opportunistic infection.

Study Subjects Evaluation

Demographic features (age, gender and smoking habits) and clinical characteristics of the subjects were recorded. Clinical symptoms (cough, sputum, fever, hemoptysis, weight loss, night sweats and lassitude) were evaluated for each participant and the total number of the present symptoms were coded as "Total Symptom Score". Bacille Calmette-Guérin (BCG) vaccine scars were counted and the Tuberculin skin test (TST) was performed on all recruited participants. The TST was executed by the intradermal placement of mL 0.1 of 5 tuberculin units (TU) of purified protein

derivative S (PPD-S) (BB-NCI PD ltd, Sofia, Bulgaria) on the forearm. The result was reported as millimeters of induration in the transverse diameter within 48–72 hours of evaluation. The inflammatory markers erythrocyte sedimentation rate (ESR), albumin level and white blood cell (WBC) from the complete blood count (CBC) were recorded from all participants. Sputum specimens had been examined by Ziehl-Neelsen staining and cultured for *M. tuberculosis* on Löwenstein-Jensen medium. The number of acid-fast bacilli (AFB) observed was quantified according to Centers for Disease Control and Prevention guidelines (Table 1) [16]. Standard full-size posteroanterior chest-X-ray (CXR)s were performed and evaluated independently by two pulmonologists, and the final decision was established by consensus at the second reading. X-ray findings were described as normal with no lesions or abnormal according to a) Presence of one or more of infiltrates, cavitary lesion, destroyed lung; b) Extent of lesions: unilateral or bilateral. The existence of extrapulmonary involvements was also noted.

Measurement of CXCL12 (SDF-1α)

For the plasma collection, EDTA was used as an anticoagulant. After collecting blood, in 30 minutes, the plasma was removed by centrifugation at 1000 ×g for 15 minutes. Collected plasmas were stored at - 80°C until measurement. CXCL12 levels in plasma were measured by enzyme-linked immunosorbent assay (ELISA) kit (Quantikine ELISA Kit, R&D system Co. Ltd., Minneapolis, MN, USA). The assay was performed according to the manufacturer's instructions.

Table 1. Sputum smear grading, according to the number of acid-fast bacilli (AFB) visualized, for light and fluorescence microcopy [16].

Number. of AFB seen		Sputum smear grade
0		Negative
1–2 per whole smear		Doubtful positive
1-9 per		
	100 fields	1+
	10 fields	2+
	1 field	3+
9 per single field		4+

The coefficient for inter- and intra-assay variation were 9.4% and 3.4%, respectively.

Statistical Method

Statistical Package for the Social Sciences (SPSS) Version 25 (SPSS, Chicago, IL) was used for the statistical analysis of this study. Comparisons between groups were performed by the Kruskal-Wallis test for nonparametric variables and the Mann-Whitney *U* test was used for subgroup analysis. The group characteristics were compared using the "Fisher exact test" and the Chi-square test. A 95% confidence interval (CI) was used to determine the cut-off value for CXCL12 (SDF-1α) level. The upper limit of 95% CI, that is, mean + 2SD of CXCL12 (SDF-1α) of HC group was taken as the cut-off value. Continuous test variable of CXCL12 (SDF-1α) was converted to dichotomous state variable on the basis of the cutoff value and the capacity of serum CXCL12 (SDF-1α) cutoff value in predicting presence of active TB infection was analyzed by constructing ROC (receiver operating characteristic)

Table 2. Comparison of demographic characteristics of study groups.

	aTbc	cTbc	CC	inf COPD	HC		Total
	n = 30	n = 15	n = 15	n = 15	n = 15	p value	n = 90
Age, years median (min-max)	32.5 (16-60)	30 (17-68)	34 (26-55)	72 (52-80)	28 (18-35)	< 0.0001	33 (16-80)
Gender, n (%)						0.923	
Female	18 (60)	9 (60)	9 (60)	7 (46.7)	9 (60)		52 (57.8)
Male	12 (40)	6 (40)	6 (40)	8 (53.3)	6 (40)		38 (42.2)
Smokers							
n (%)	16 (53.3)	6 (40)	6 (40)	8 (53.3)	7 (46.7)	0.863	43 (47.8)
pack year, median (min-max)	1 (0-50)	0 (0-45)	0(0-25)	20 (0-150)	0 (0-18)	0.279	0 (0-150)
BCG scars, n						0.001	
0	2 (6.7)	0 (0)	0 (0)	6 (40)	0 (0)		8 (8.9)
1	15 (50)	8 (53.3)	5 (33.3)	8 (53.3)	7 (46.7)		43 (47.8)
2	11 (36.7)	6 (40)	10 (66.7)	1 (6.7)	8 (53.3)		36 (40)
3	2 (6.7)	1 (6.7)	0 (0)	0 (0)	0 (0)		3 (3.3)
TST							
mm, median (min-max)	11.5 (0-20)	12 (1-20)	14(4-34)	8 (0-12)	11 (0-20)	0.034	11 (0-34)
TST- positive, n (%)	4 (13.3)	4 (28.6)	7 (46.7)	3 (20)	4 (26.7)	0.178	22 (24.7)
TST-negative, n (%)	26 (86.7)	10 (71.4)	8 (53.3)	12 (80)	11 (73.3)		67 (75.3)

curve and measuring the area under the curve (AUC). Positive predictive value (PPV) and negative predictive value (NPV) were calculated for cutoff value. A p-value < 0.05 was considered statistically significant.

Results

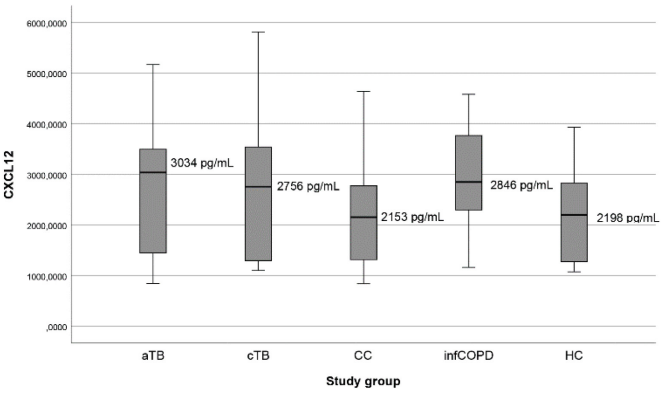
Study Population

The study included 90 patients (52 females, 57.8%) with a median age of 33 (min-max:16-80). Forty-three (47.8%) of the patients were current cigarette smokers. The study groups were comparable in the term of demographic characteristics except for age, ESR and BCG scar presence ($p < 0.0001$, $p < 0.0001$ and 0.001, respectively) (Table 2). Also, Table 3 summarizes the clinical and radiological findings of all participants.

Analysis of CXCL12 (SDF-1α) Levels According to Study Groups

Median CXCL12 (SDF-1α) level was 2658 pg/mL (min-max; 839-5813) in overall study group. Although the patients in aTB group had the highest median CXCL12 (SDF-1α) levels (3034 pg/mL), it was statistically non-significant both in group and subgroup analysis ($p = 0.3$). Median CXCL12 (SDF-1α) levels were 2756 pg/mL in cTB group, 2153 pg/mL in CC group, 2846 pg/mL in infCOPD group and 2198 pg/mL in HC group (Figure 1).

Figure 1. Plasma levels of CXCL12 (SDF-1α) chemokine in participants from all study groups. In the box plots, horizontal lines represent medians (25th and 75th percentiles) and error bars represent the 10th and 90th percentiles. Median CXCL12 (SDF-1α) level of each group is given by its horizontal line.



The cut-off value for CXCL12 (SDF-1α) level was found to be 2835 pg/mL. When 2835 pg/mL was defined as cut-off value for CXCL12 (SDF-1α) 39 (43.3%) patients had increased CXCL12 (SDF-1α) levels. Although the rate of patients with increased CXCL12 (SDF-1α) was highest in aTB group, it was statistically non-significant ($p = 0.09$). In the subgroup analysis, increased CXCL12 (SDF-1α) rate was significantly more in aTB group when it was compared with CC ($p = 0.02$) and HC ($p = 0.05$) groups (Figure 2).

Table 3. Comparison of clinical and radiological findings of study groups.

	aTbc	cTbc	CC	inf COPD	HC	
	n = 30	n = 15	n = 15	n = 15	n = 15	p value
Symptoms, (n, %)						
Cough	28 (93.3)	1 (6.7)	0	14 (93.3)	0	< 0.0001
Sputum	18 (60)	0	0	10 (66.7)	0	< 0.0001
Fever	12 (40)	0	0	5 (33.3)	0	< 0.0001
Hemoptysis	2 (6.7)	0	0	0	0	0.394
Weight loss	16 (53.3)	0	0	1 (6.7)	0	< 0.0001
Night sweats	16 (53.3)	0	0	1 (6.7)	0	< 0.0001
Lassitude	11 (36.7)	0	0	1 (6.7)	0	< 0.0001
Total symp. score, median (min-max)	3 (1-6)	0 (0-1)	0 (0)	2 (0-5)	0 (0)	< 0.0001
ESR, mm/hr, median (min-max)	65 (30-80)	10 (2-35)	10 (3-12)	20 (2-45)	3 (1-10)	< 0.0001
AFP smear grade						< 0.0001
+ (n, %)	11(36.7)	0	0	0	0	
++ (n, %)	4 (13.3)	0	0	0	0	
+++ (n, %)	3 (10)	0	0	0	0	
++++ (n, %)	12 (40)	0	0	0	0	
Chest X-ray findings						< 0.0001
Infiltrate (n, %)	4 (13.3)	0	0	8 (53.3)	0	
Cavitation +Inf (n, %)	25 (83.3)	0	0	0	0	
Destroyed lung + Cav +Inf (n, %)	1 (3.3)	0	0	0	0	
Involvement of chest X-ray						< 0.0001
Unilateral (n, %)	13 (43.3)	0	0	4 (26.7)	0	
Bilateral (n, %)	17 (56.7)	0	0	4 (26.7)	0	

Figure 2. Increased CXCL12 rates in all five categories of participants. Although the rate of patients with elevated CXCL12 was highest in aTB group, it was statistically non-significant (p = 0.09). In the subgroup analysis, increased CXCL12 rate was significantly more in aTB group when it was compared with CC (p = 0.02) and HC (p = 0.05) groups. aTB, active TB; cTB, cured TB; CC, close contacts of aTB patients; infCOPD, chronic obstructive pulmonary disease (COPD) with active nonspecific pulmonary infection; HC, healthy controls.

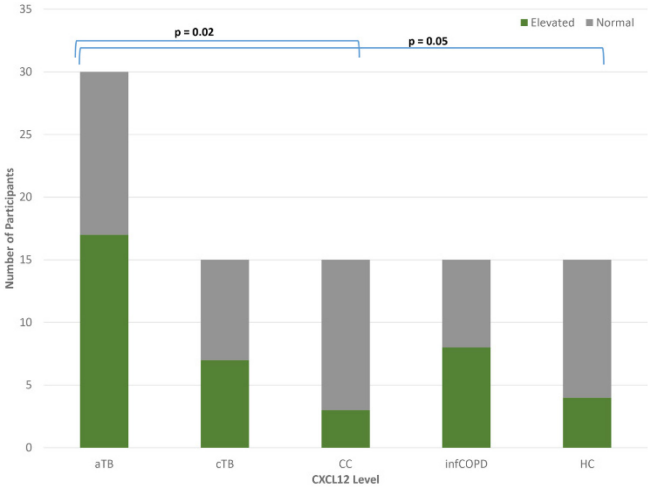


Figure 3. Receiver Operating Characteristic (ROC) Curve was built to evaluate overall diagnostic value of CXCL12 (SDF-1α) for active TB infection.

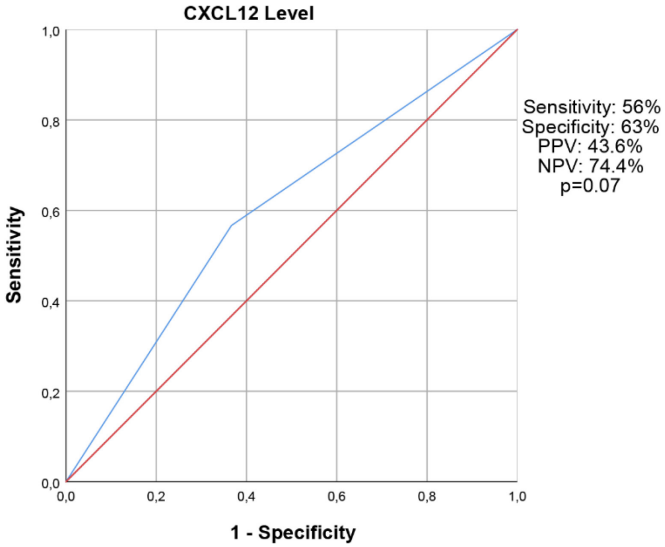


Table 4. Comparison of demographic, biochemical and radiologic findings in study participants according to the presence of increased CXCL12 (SDF-1α) levels.

	Increased CXCL12 n = 39	Normal CXCL12 n = 51	p value
TST			0.699
TST- positive, n (%)	9 (23.1)	13 (25.5)	
TST-negative, n (%)	29 (74.4)	38 (74.5)	
Symptoms, (n, %)			
Cough	23 (59)	20 (39.2)	0.063
Sputum	19 (48.7)	9 (17.6)	0.002
Fever	12 (30.8)	5 (9.8)	0.012
Hemoptysis	1 (2.6)	1 (2)	0.847
Weight loss	9 (23.1)	8 (15.7)	0.375
Night sweats	8 (20.5)	9 (17.6)	0.731
Lassitude	6 (15.4)	6 (11.8)	0.617
Total symp. score, Median (min-max)	2 (0-6)	0 (0-5)	0.019
AFP smear grade			0.057
+ (n, %)	3 (17.6)	8 (61.5)	
++ (n, %)	2 (11.8)	2 (15.4)	
+++ (n, %)	2 (11.8)	1 (7.7)	
++++ (n, %)	10 (58.8)	2 (15.4)	
Chest X-ray Findings			0.032
Infiltrate (n, %)	9 (23.1)	3 (5.9)	
Cavitation +Inf (n, %)	12 (30.8)	13 (25.5)	
Destroyed lung + Cav +Inf (n, %)	1 (2.6)	0	
Involvement of Chest X-ray			0.010
Unilateral (n, %)	7 (17.9)	10 (19.6)	
Bilateral (n, %)	15 (38.5)	6 (11.8)	
ESR, mm/hr, Median (min-max)	30 (2-80)	10 (1-80)	0.095

Receiver Operating Characteristic (ROC) Curve Analysis of CXCL12 (SDF-1α) levels to Predict Active Tuberculosis Infection

A ROC curve was built to evaluate the overall diagnostic value of CXCL12 (SDF-1α) for active TB infection. This analysis showed a statistically quasi-significant area under the curve of 0.600, with a standard error of 0.05, a specificity of 63% and sensitivity of 56% (p = 0.07, 95% confidence interval 0.491–0.702). PPV and NPV were 43.6% and 74.5%, respectively (Figure 3).

Comparison of Demographic, Clinical, Biochemical and Radiological Findings of Study Participants According to Increased CXCL12 (SDF-1α) Levels

Clinical, biochemical, radiological and demographic characteristics were compared between patients according to the presence of increased CXCL12 (SDF-1α) levels (Table 4). No statistical difference was shown within the groups for age, gender, smoke, WBC and albumin level (*p-value* is 0.401, 0.135, 0.876, 0.818 and 0.858, respectively). Presence of fever and sputum, also higher total symptom scores, more severe CXR infiltration and involvement were significantly seen more in patients with increased CXCL12 (SDF-1α) levels (*p-value* is 0.012, 0.002, 0.019, 0.032 and 0.010, respectively).

Active Infection and CXCL12 (SDF-1α) levels

While patients were divided into two groups according to the presence of active infection, patients with an active infection (aTB and infCOPD) had higher increased CXCL12 (SDF-1α) rates (64.1% vs. 35.9%) (p = 0.01).

Prediction of independent factors for increased CXCL12 (SDF-1α) levels

When binary logistic regression analysis was carried out, the results showed that AFP smear grade, the radiological severity of chest x-ray findings and extend of chest x-ray involvement were independent factors for increased CXCL12 levels (OR:2.973, 95%CI [1.213-7.286], OR:0.199, 95%CI [0.041-0.972], OR:21.222, 95%CI [1.569-287.101], respectively) (Table 5).

Discussion

New accurate, rapid and low-cost diagnostic tests are needed in order to decrease diagnostic errors and to allow early detection of TB. Implementation of these new tests may help to reduce the risks of tuberculosis transmission, poor treatment outcomes, prevention of adverse sequelae and mortality rates [2].

Infection of human cells with *M. tuberculosis* induces the protective immune response to release multiple factors including the production of specific cytokines and chemokines [17]. Various type of chemokines has been studied in different clinical and animal models of TB infection. One of these chemokines, CXCL12 (SDF-1α), not only participates in hematopoietic stem cell mobilization and migration of leukocytes such as early B cell progenitors, B/T cell subpopulations, monocytes, megakaryocytes and CD34-positive stem cells but also plays critical roles in organogenesis and wound healing [14,18]. CXCL12 has two receptors: CXCR4 and CXCR7 [19]. CXCR4 is the only inflammatory chemokine receptor that has only one ligand, CXCL12. In addition to this, it is the only chemokine receptor of which knockout mice die perinatally. Similar to that of CXCL12 knockout mice also die perinatally, underlining the crucial role of this axis [12]. CXCL12 is normally highly expressed in the bone marrow and acts as a retention factor for neutrophils. A decrease in its concentration in the bone marrow is seen in the face of inflammation, while an increase at the site of inflammation is seen, causing PMNs to enter the circulation and to migrate to the inflammation site [19]. CXCL12 chemokine was found to be associated with pulmonary granuloma formation, organized lymphoid structure, T lymphocytes and neutrophils prevalence and may be a potential biological marker for tuberculosis diagnosis [20].

Table 5. Binary logistic regression analysis results to predict independent factors for increased CXCL12 levels.

	OR	p	95% CI
Cough	0.748	0.784	0.094-5.949
Sputum	0.178	0.139	0.018-1.748
Fever	0.184	0.117	0.022-1.527
Total symptom score	0.467	0.197	0.147-1.485
AFP smear grade	2.973	0.017	1.213-7.286
Chest X-ray findings	0.199	0.046	0.041-0.972
Involvement of chest X-ray	21.222	0.022	1.569-287.101
ESR	1.004	0.858	0.964-1.046

There is limited and contradictory data in the literature concerning the relationship of CXCL12 (SDF-1α) with *M. tuberculosis* related diseases. These discrepancies are probably because different models and parameters were used in these different studies. Therefore, we conducted this study with diverse groups of patients in order to have a chance to evaluate the CXCL12 levels with the different aspects of the inflammation. We tried to find out the answer to the question; "Is it the infection or is it only the TB infection/disease itself that increases CXCL12 level?".

In the present study, although the plasma levels of CXCL12 (SDF-1α) were highest in the aTB group, no significant difference was seen between groups. On the other hand, when a cut-off level for CXCL12 was determined, that will optimize the detection of positive test results, we noticed that increased CXCL12 rate was significantly more in aTB group than CC and HC. In order to evaluate the overall diagnostic value of CXCL12 for aTB, a ROC curve was built. The sensitivity and specificity were found to be 0.56 and 0.63, respectively, indicating that this test may not be feasible to use in diagnosing aTB.

When we divided the participant groups into two, according to the presence of active infection (aTB and infCOPD vs. cTB, CC and HC), patients with active infection had statistically significant higher increased CXCL12 rates. Also, the indicators of an active infection, like fever and sputum, higher total symptom scores, more severe CXR infiltration and involvement were significantly seen more in patients with increased CXCL12 (SDF-1α) levels.

The major finding of this study was that active infection (aTB and infCOPD) itself; regardless of the cause but correlated with the bacterial load and also with the severity of CXR findings; is the factor responsible for increased CXCL12 levels. We propose two mechanisms that likely contribute to this condition: (i) Since CXCL12 is one of the most efficacious chemoattractants for the recruitment of inflammatory cells to the site of inflammation caused by the infection, excessive amounts of CXCL12 is produced at the site of infection. It has been reported that CXCL12 (SDF-1α) was elevated in the plasma of aTB and pleural effusions of TB pleurisy in several studies [4,14,15]. Moreover, CXCL12 has been shown to be associated with infectious diseases other than TB such as human immunodeficiency virus (HIV) infection, viral hepatitis and sepsis [21-23]. (ii) It has been found that fibrocyte differentiation and proliferation capacity is increased in patients with aTB and also in acute exacerbations of COPD, then from normal subjects. CXCL12 participates in mobilizing bone marrow-derived stem cells, via its receptor CXCR4, including fibrocytes. CXCL12/CXCR4 axis is involved in fibrocyte recruitment during aTB and acute exacerbations of COPD [24,25]. Therefore, CXCL12 (SDF-1α) cannot be used as a diagnostic marker but maybe as a prognostic factor for aTB which needs further evaluation.

Although we documented such association, some limitations must be recognized. An important limitation is the small size of our sample that could obviously affect the potency of our analysis. However, we think that it points to the need for further investigation to confirm our data, and our observations can represent the foundation for larger trials aimed to evaluate the prognostic role of CXCL12 (easily measurable by ELISA test) in aTB. Another limitation is that infCOPD group's age was older than the other groups, which is an expected result. And also, this age inconsistency resulted a mismatch in the vaccination schedule and BCG scar presence. However, these might not affect the results, since they were not found to be correlated with CXCL12 levels.

Conclusions

In conclusion, our study demonstrates that CXCL12 (SDF-1α) may be a representative of active infection regardless of the cause but correlated with the severity of the disease; enabling this test to be used as a prognostic factor rather than a diagnostic test for aTB. Nevertheless, further studies are required to assess the robustness of our findings.

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Corresponding author

Ilkay Koca Kalkan, MD
Atatürk Chest Disease and Thoracic Surgery Training and Research Hospital, Department of Allergy and Clinical Immunology
Sanatoryum Caddesi, 06280, Kecioren, Ankara, Turkey
Phone: +905053117710
Fax: +90312 5677000
E-mail address: ilkay.koca@gmail.com

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